

CLAIMS

1. A method for separating proteins comprising the steps of:

(a) adsorbing a target protein on an ion exchanger by allowing a
5 sample containing the target protein to contact the ion exchanger under
a first condition at high ion strength and at a pH outside of the
vicinity of the isoelectric point of the target protein; and

(b) eluting the component adsorbed on the ion exchanger under a
second condition at lower ion strength than in the first condition, and
10 at a pH closer to the isoelectric point side of the protein in the
first condition.

2. The method for separating proteins according to Claim 1
comprising the step of using a buffer solution with a concentration of
15 0.05 M or more in the first condition.

3. The method for separating proteins according to Claim 1
comprising the steps of using a high concentration of the buffer
solution comprising a combination of a weak acid and weak base in the
20 first condition.

4. A method for separating proteins comprising the steps of:

(a) adsorbing a target protein on an ion exchanger by allowing a
sample containing the target protein to contact the ion exchanger under
25 a first condition at high ion strength and at a pH outside of the
vicinity of an isoelectric point of the target protein; and

(b) eluting the component adsorbed on the ion exchanger under a
second condition at ion strength equal to or lower than in the first
condition, and at a pH closer to the isoelectric point side of the
30 protein in the first condition.

5. The method for separating proteins according to Claim 1
comprising the following step interposed between step (a) and step (b):

(c) washing the ion exchanger under a condition not eluting the
35 target protein adsorbed on the ion exchanger.

6. The method for separating proteins according to Claim 5,
wherein step (c) is applied under a substantially the same condition as
in the first condition.

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7. The method for separating proteins according to Claim 1,
wherein

the pH in the first condition is lower than the isoelectric point
of the target protein,

10 the ion exchanger is a cation exchanger, and

the pH in the second condition is in the vicinity of or higher
than the isoelectric point of the target protein.

8. The method for separating proteins according to Claim 1,
15 wherein

the pH in the first condition is higher than the isoelectric
point of the target protein,

the ion exchanger is an anion exchanger, and

20 the pH in the second condition is in the vicinity of or lower
than the pH corresponding to the isoelectric point of the target
protein.

9. The method for separating proteins according to Claim 1
comprising the step of using a tris-succinate buffer in the first
25 condition.

10. The method for separating proteins according to Claim 1,
wherein the second condition comprises the step of using a buffer
solution comprising a combination of the same acid and same base as in
30 the buffer solution used in the first condition.

11. The method for separating proteins according to Claim 1,
wherein the second condition comprises the step of using a buffer
solution having a pH in the vicinity of the isoelectric point of the
35 target protein.

12. The method for separating proteins according to Claim 1,
wherein

the sample contains a plurality of target proteins, and
5 step (b) comprises the step of continuously eluting the target
proteins under a solvent condition corresponding to the isoelectric
point of each protein.

13. The method for separating proteins according to Claim 1,
10 wherein the protein is a glycoprotein.